

WHAT IS CLAIMED IS:

- 5 1. A conjugate comprising at least one non-polypeptide moiety covalently attached to a polypeptide, wherein the amino acid sequence of the polypeptide differs from that of wild-type FVII or FVIIa shown in SEQ ID NO:1 in that at least one amino acid residue comprising an attachment group for said non-polypeptide moiety has been introduced or removed.
- 10 2. The conjugate according to claim 1, wherein the attachment group is selected from the group consisting of lysine, cysteine, aspartic acid and glutamic acid.
3. The conjugate according to claim 2, wherein the attachment group is lysine.
4. The conjugate according to claim 3, wherein the amino acid sequence of the polypeptide differs from SEQ ID NO:1 in that at least one lysine residue has been removed.
- 15 5. The conjugate according to claim 4, wherein the lysine residue has been removed by substitution.
6. The conjugate according to claim 5, wherein the removed lysine residue is selected from the group consisting of K18, K32, K38, K62, K85, K109, K137, K143, K148, K157, K161, K197, K199, K316, K337, K341, K389 and combinations thereof.
- 20 7. The conjugate according to claim 6, wherein the lysine residue is selected from the group consisting of K18, K62, K85, K197, K341 and combinations thereof.
8. The conjugate according to claim 7, wherein the lysine residue is substituted with an amino acid residue selected from the group consisting of R, Q, N and H.
- 25 9. The conjugate according to claim 8, wherein the lysine residue is substituted with R.
10. The conjugate according to claim 3, wherein the amino acid sequence of the polypeptide differs from SEQ ID NO:1 in that at least one lysine residue has been introduced.

5 **11.** The conjugate according to claim 10, wherein the lysine residue has been introduced by substitution.

12. The conjugate according to claim 11, wherein the substitution is selected from the group consisting of I42K, Y44K, L288K, D289K, R290K, G291K, A292K, T293K, Q313K, S314K, R315K, V317K, L390K, M391K, R392K, S393K, E394K, P395K, R396K,
10 P397K, G398K, V399K, L400K, L401K, R402K, A403K, P404K, F405K and combinations thereof.

13. The conjugate according to claim 12, wherein the substitution is selected from the group consisting of R290K, R315K, R392K, R396K, R402K and combinations thereof.

15 **14.** The conjugate according to claim 3, wherein at least one lysine residue has been removed and at least one lysine residue has been introduced.

15. The conjugate according to claim 2, wherein the attachment group is cysteine.

16. The conjugate according to claim 15, wherein the cysteine residue has been introduced by substitution.

20 **17.** The conjugate according to claim 16, wherein the substitution is selected from the group consisting of I30C, K32C, D33C, A34C, T37C, K38C, W41C, Y44C, S45C, D46C, L141C, E142C, K143C, R144C, L288C, D289C, R290C, G291C, A292C, S314C, R315C, K316C, V317C, L390C, M391C, R392C, S393C, E394C, P395C, R396C, P397C, G398C, V399C, L401C, R402C, A403C, P404C and combinations thereof.

25 **18.** The conjugate according to claim 17, wherein the substitution is selected from the group consisting of K32C, Y44C, K143C, R290C, R315C, K341C, R392C, R396C, R402C and combinations thereof.

19. The conjugate according to claim 2, wherein the attachment group is aspartic acid and/or glutamic acid

5 **20.** The conjugate according to claim 19, wherein the amino acid sequence of the polypeptide differs from SEQ ID NO:1 in that at least one aspartic acid residue and/or glutamic acid residue has been introduced.

21. The conjugate according to claim 20, wherein the aspartic acid residue and/or the glutamic acid residue has been introduced by substitution.

10 **22.** The conjugate according to claim 21, wherein the substitution is selected from the group consisting of I30D/E, K32D/E, A34D/E, T37D/E, K38D/E, W41D/E, Y44D/E, S45D/E, D46C, L141D/E, E142D/E, K143D/E, R144D/E, L288D/E, R290D/E, G291D/E, A292D/E, Q313D/E, S314D/E, R315D/E, K316D/E, V317D/E, L390D/E, M391D/E, R392D/E, S393D/E, P395D/E, R396D/E, P397D/E, G398D/E, V399D/E, L401D/E, R402D/E, A403D/E, P404D/E, and combinations thereof.

15 **23.** The conjugate according to claim 22, wherein the substitution is selected from the group consisting of K32D/E, Y44D/E, K143D/E, R290D/E, R315D/E, K341D/E, R392D/E, R396D/E, R402D/E and combinations thereof.

20 **24.** The conjugate according to claim 19, wherein the amino acid sequence of the polypeptide differs from SEQ ID NO:1 in that at least one aspartic acid residue and/or glutamic acid residue has been removed.

25 **25.** The conjugate according to claim 24, wherein the aspartic acid residue and/or glutamic acid residue has been removed by substitution.

26. The conjugate according to claim 25, wherein the substitution is selected from the group consisting of D33, D46, D48, E77, E82, D86, D87, E94, E99, D104, E116, D123, E132, E142, E163, D196, E210, D212, E215, D217, D219, E220, D256, E265, E270, D289, E296, D309, D319, E325, D334, D338, D343, E385, E394 and combinations thereof.

30 **27.** The conjugate according to claim 19, wherein at least one aspartic acid and/or glutamic acid residue has been introduced and at least one aspartic acid and/or glutamic acid residue has been removed.

5 28. The conjugate according to claim 1, wherein the non-polypeptide moiety is a polymer molecule.

 29. The conjugate according to claim 28, wherein the polymer molecule is selected from the group consisting of natural or synthetic homopolymers and heteropolymers.

10 30. The conjugate according to claim 29, wherein the polymer molecule is a synthetic homo-polymer or hetero-polymer selected from the group consisting of linear and branched polyethylene glycol, polyvinyl alcohol (PVA), polycarboxylic acids and poly-(vinylpyrrolidone).

 31. The conjugate according to claim 30, wherein the polymer is a linear or branched polyethylene glycol.

15 32. The conjugate according to claim 31, wherein the polyethylene glycol has a molecular weight of about 300 to 100,000 Da.

 33. The conjugate according to claim 1, wherein the polypeptide differs in 1-15 amino acid residues from the amino acid sequence shown in SEQ ID NO:1.

20 34. The conjugate according to claim 1, wherein said non-polypeptide moiety is a sugar moiety and wherein said attachment group is an attachment group for a sugar moiety.

 35. The conjugate according to claim 34, wherein said attachment group is a glycosylation site.

 36. The conjugate according to claim 35, wherein at least one non-naturally occurring glycosylation site has been introduced.

25 37. The conjugate according to claim 35, wherein at least one naturally occurring glycosylation site has been removed.

 38. The conjugate according to claim 35, wherein at least one non-naturally occurring glycosylation site has been introduced and at least one naturally occurring glycosylation site has been removed.

- 5 **39.** The conjugate according to claim 36, wherein the introduced glycosylation site is an *in vivo* glycosylation site.
- 40.** The conjugate according to claim 39, wherein the glycosylation site is an *in vivo* O-glycosylation site.
- 41.** The conjugate according to claim 39, wherein the glycosylation site is an *in vivo* N-glycosylation site.
- 10 **42.** The conjugate according to claim 41, wherein the amino acid sequence of the polypeptide differs from SEQ ID NO:1 in that at least one naturally occurring N-X'-X sequence is substituted with a N-X'-S or N-X'-T sequence, wherein X' is any amino acid except P, and X is any amino acid except for S and T.
- 43.** The conjugate according to claim 41, wherein the amino acid sequence of the polypeptide differs from SEQ ID NO:1 in that at least one naturally occurring X-X'-S or X-X'-T sequence naturally present in SEQ ID NO:1 is substituted with a N-X'-S or a N-X'-T sequence, wherein X' is any amino acid except P, and X is any amino acid except for N.
- 44.** The conjugate according to claims 42 or 43, wherein the substitution is selected from the group consisting of F4S/T, P10N, Q21N, W41N, S43N, A51N, G58N, L65N, G59S/T, E82S/T, N95S/T, G97S/T, Y101N, D104N, T106N, K109N, G117N, G124N, S126N, T128N, A175S/T, G179N, I186S/T, V188N, R202S/T, I205S/T, D212N, E220N, I230N, P231N, P236N, G237N, V253N, E265N, T267N, E270N, R277N, L280N, G291N, P303S/T, L305N, Q312N, G318N, G331N, D334N, K337N, G342N, H348N, R353N, Y357N, I361N, V376N, R379N, M391N, and combinations thereof.
- 25 **45.** The conjugate according to claim 44, wherein the substitution is selected from the group consisting of F4S/T, P10N, Q21N, W41N, A51N, G58N, G59S/T, N95S/T, G97S/T, Y101N, D104N, T106N, K109N, G117N, G124N, S126N, T128N, A175S/T, I186S/T, V188N, R202S/T, I205S/T, D212N, E220N, V253N, E265N, T267N, E270N, L280N, G291N, P303S/T, G318N, G331N, D334N, K337N, R353N, Y357N, M391N, and combinations thereof.
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5 46. The conjugate according to claim 41, wherein the amino acid sequence of the polypeptide differs from SEQ ID NO:1 in that at least one K-X'-X sequence or R-X'-X sequence naturally present in SEQ ID NO:1 is substituted with a N-X'-S or a N-X'-T sequence, wherein X' is any amino acid except P, and X is any amino acid except for S and T.

10 47. The conjugate according to claim 46, wherein the substitution is selected from the group consisting of K32N+A34S/T, K38N+F40S/T, K62N+Q64S/T, K85N+D87S/T, K137N+P139S/T, K143N+N145S/T, K148N+Q149S/T, K161N+E163S/T, K197N+K199S/T, K199N+W201S/T, K316N+G318S/T, K337N, K341N+D343S/T, K389N+M391S/T and combinations thereof.

15 48. The conjugate according to claim 41, wherein a non-naturally occurring *in vivo* N-glycosylation site is introduced by substitution, the substitution being selected from the group consisting of K32N+A34S/T, F31N+D33S/T, I30N+K32S/T, A34N+R36S/T, K38N+F40S/T, T37N+L39S/T, R36N+K38S/T, L39N+W41S/T, F40N+I42S/T, W41N, I42N+Y44S/T, S43N, Y44N+D46S/T, S45N+G47S/T, D46N+D48S/T, G47N+Q49S/T, K143N+N145S/T, E142N+R144S/T, L141N+K143S/T, I140N+E142S/T, R144N+A146S/T, 20 A146N+K148S/T, S147N+P149S/T, R290N+A292S/T, D289N+G291S/T, L288N+R290S/T, L287N+D289S/T, G291N, A292N+A294S/T, T293N+L295S/T, R315N+V317S/T, S314N+K316S/T, Q313N+R315S/T, Q312N, K316N+G318S/T, V317N+D319S/T, G318N, K341N+D343S/T, S339N+K341S/T, G342N, D343N+G345S/T, R392N+E394S/T, M391N, L390N+R392S/T, K389N+M391S/T, S393N+P395S/T, E394N+R396S/T, P395N+P397S/T, 25 R396N+G398S/T, P397N+V399S/T, G398N+L400S/T, V399N+L401S/T, L400N+R402S/T, L401N+A403S/T, R402N+P404S/T, A403N+F405S/T, P404N+P406S/T, K143N+N145S/T+ R315N+V317S/T and combinations thereof.

30 49. The conjugate according to claim 48, wherein the substitution is selected from the group consisting of K32N+A34S/T, K38N+F40S/T, Y44N+D46S/T, K143N+N145S/T, R290N+A292S/T, K341N+D343S/T, R392N+E394S/T, R396N+G398S/T, R402N+P404S/T, K143N+N145S/T+ R315N+V317S/T and combinations thereof.

50. The conjugate according to claim 49, wherein the substitution is selected from the group consisting of K32N+A34T, K38N+F40T, Y44N+D46T, K143N+N145T,

5 R290N+A292T, K341N+D343T, R392N+E394T, R396N+G398T, R402N+P404T,
K143N+N145T+ R315N+V317T and combinations thereof.

51. The conjugate according to claim 34, wherein the polypeptide differs in 1-15
amino acid residues from the amino acid sequence shown in SEQ ID NO:1.

10 52. The conjugate according to claim 39, wherein two or more *in vivo*
glycosylation sites have been introduced.

53. The conjugate according to claim 34, which further comprises at least one
non-polypeptide moiety covalently attached to an amino acid residue of the polypeptide, wherein
the non-polypeptide moiety is different from a sugar moiety.

15 54. The conjugate according to claim 53, wherein the non-polypeptide moiety is
a polymer molecule as defined in claim 29.

55. The conjugate according to claim 1, wherein the conjugate has a molecular
weight of at least 67 kDa.

20 56. The conjugate according to claim 1, wherein the conjugate has been
inactivated.

57. A polypeptide having an amino acid sequence as defined in claim 1.

58. A nucleotide sequence encoding the polypeptide of claim 57.

59. An expression vector harbouring the nucleotide sequence of claim 58.

60. A host cell comprising a nucleotide sequence according to claim 58 or an
expression vector according to claim 59.

25 61. The host cell according to claim 60, wherein the host cell is a
gamma-carboxylating cell capable of *in vivo* glycosylation.

62. A method for producing a conjugate as defined in claim 1, comprising

5 culturing a host cell as defined in claim 66 under conditions conducive for the expression of the polypeptide, and recovering the polypeptide, wherein a) the polypeptide comprises at least one N- or O-glycosylation site and the host cell is a eukaryotic host cell capable of *in vivo* glycosylation, and/or b) the polypeptide is subjected to conjugation to a non-polypeptide moiety *in vitro*.

10 **63.** A composition comprising a conjugate as defined in claim 1 and a pharmaceutical acceptable carrier or excipient.

64. A method for treating a mammal having a FVIIa/TF-related disease or disorder, comprising administering to a mammal in need thereof an effective amount of a conjugate as defined in claim 1.

15 **65.** The method according to claim 64, wherein the disease or disorder is selected from the group consisting of diseases where increased clot formation is desirable.

66. A method for treating a mammal having a FVIIa/TF-related disease or disorder, comprising administering to a mammal in need thereof an effective amount of a conjugate as defined in claim 56.

20 **67.** The method according to claim 66, wherein the disease or disorder is selected from the group consisting of diseases where decreased clot formation is desirable.

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